CLAIMS

What is claimed is:

1. A method for high level expression of recombinant forms of the Receptor for Advanced Glycated Endproducts (RAGE) or fragments thereof comprising:

(a) subcloning a nucleotide sequence encoding RAGE or a fragment thereof into a virus;

(b) preparing a high-titer stock of recombinant virus; and

(c) infecting host cells with the high-titer recombinant virus under conditions such that pre-determined levels of RAGE or a fragment thereof is produced, wherein said pre-determined levels of RAGE comprises at least 25 mg recombinant protein per liter of culture.

2. The method of claim 1, further comprising a yield of recombinant RAGE polypeptide of more than 50 mg per liter of culture.

3. The method of claim 1, further comprising a yield of recombinant RAGE polypeptide of more than 100 mg per liter of culture.

4. The method of claim 1, further comprising a yield of recombinant RAGE polypeptide of more than 250 mg per liter of culture.

5. The method of claim 1, wherein the virus comprises the *Autographa californica* nuclear polyhedrosis virus.

6. The method of claim 1, wherein the host cells comprise insect cells such as Sf9 or Sf21 cells.

7. The method of claim 1, wherein the recombinant RAGE protein or fragment thereof is purified from the insect media using Sepharose.

8. The method of claim 1, further comprising infecting insect cells at a multiplicity of infection (MOI) of less than 1, and incubating the insect cell culture at a temperature of about 26-28°C for 3-7 days to prepare high titer virus stock.

9. The method of claim 8, wherein the innoculum used to prepare the high titer stock comprises a multiplicity of infection (MOI) of 0.01 to 1.0.

10. The method of claim 8, wherein the innoculum used to prepare the high titer stock comprises a multiplicity of infection (MOI) of 0.05 to 0.5.

11. The method of claim 8, wherein the innoculum used to prepare the high titer stock comprises a multiplicity of infection (MOI) of 0.1 to 0.2.

12. The method of claim 1, wherein the nucleotide sequence encoding RAGE comprises SEQ ID NO: 1, or a sequence substantially homologous thereto.

13. The method of claim 1, wherein the fragment of RAGE subcloned for expression is the soluble, extracellular portion of RAGE (sRAGE), as encoded by the nucleic acid sequence SEQ ID NO: 2, or a sequence substantially homologous thereto.

14. The method of claim 1, wherein the fragment of RAGE subcloned for expression is the V-domain of RAGE, as encoded by the nucleic acid sequence SEQ ID NO: 4, or a sequence substantially homologous thereto.

15. The method of claim 1, wherein infecting host cells under conditions such that predetermined levels of RAGE or a fragment thereof is produced comprises the steps of:

initiating cultures of insect cells at a low density;

growing the insect cells to a preset final density;

adding the high titer virus at a MOI of less than 30; and

incubating infected cells under conditions such that a predetermined level of RAGE or a fragment thereof is produced.

16. The method of claim 15, wherein the step of infecting cells at a low density comprises cells having an initial density of no more than 0.5 x 10^6 cells per ml.

17. The method of claim 15, further comprising growing the cells from an initial density of less than 0.5 x 10^6 cells per ml to a final density comprising 1 to 20 x 10^6 cells per ml.

18. The method of claim 1, wherein the cells are grown under conditions comprising a pre-set doubling time and viability.

19. The method of claim 18, wherein the rate of cell growth comprises a doubling rate of 10-35 hours.

20. The method of claim 18, wherein the rate of cell growth comprises a doubling rate of 15-30 hours.

21. The method of claim 18, wherein the rate of cell growth comprises a doubling rate of 18-26 hours.

22. The method of claim 1, wherein the doubling time comprises conditions such that cell viability is greater than 90%.

23. Insect cells producing recombinant RAGE or a fragment thereof according to claim 1.

24. Recombinant RAGE produced by the method of claim 1.

25. A method for high level expression of recombinant human Receptor for Advanced Glycated Endproducts (RAGE) comprising:

(a) preparing recombinant virus comprising a nucleotide sequence encoding RAGE or a fragment thereof subcloned into the *Autographa californica* nuclear polyhedrosis virus;

(b) preparing a high-titer stock of the recombinant virus;

(c) initiating cultures of insect cells at an initial density of less than 0.5 $x10^6$ cells per ml;

(d) growing the insect cells until the cell density comprises 1 to 20 x 10^6 cells per ml;

(e) adding virus from step (b) at a MOI of less than 30 to the insect cells from step (d) for large scale expression; and

(f) incubating the infected culture at about 26-28 °C under conditions such that a predetermined level of RAGE or a fragment thereof is produced.

26. The method of claim 25, further comprising a yield of recombinant protein of at least 25 mg per liter of culture.

27. The method of claim 25, further comprising a yield of recombinant protein of more than 100 mg per liter of culture.

28. The method of claim 25, further comprising a yield of recombinant protein of more than 250 mg per liter of culture.

29. The method of claim 25, wherein the recombinant RAGE or fragment thereof is purified from the insect media using Sepharose.

30. The method of claim 25, wherein the nucleotide sequence encoding RAGE comprises SEQ ID NO: 1, or a sequence substantially homologous thereto.

31. The method of claim 25, wherein the fragment of RAGE subcloned for expression is the soluble, extracellular portion of RAGE (sRAGE), as defined by the nucleic acid sequence SEQ ID NO: 2, or a sequence substantially homologous thereto.

32. The method of claim 25, wherein the fragment of RAGE is the V-domain of RAGE, as defined by the nucleic acid sequence SEQ ID NO: 4, or a sequence substantially homologous thereto.

33. The method of claim 25, wherein the step of preparing the recombinant virus stock comprises infecting insect cells at a multiplicity of infection (MOI) of less than 1, and incubating the insect cell culture at a temperature ranging from about 26-28°C for 3-7 days to prepare a high titer virus stock.

34. The method of claim 33, wherein the multiplicity of infection (MOI) used to prepare the high titer virus stock ranges from 0.01 to 1.0.

35. The method of claim 33, wherein the multiplicity of infection (MOI) used to prepare the high titer virus stock ranges from 0.05 to 0.5.

36. The method of claim 33, wherein the multiplicity of infection (MOI) used to prepare the high titer virus stock ranges from 0.1 to 0.2.

37. The method of claim 25, wherein the cells infected used for large scale expression are grown under conditions comprising a pre-set doubling time and viability.

38. The method of claim 37, wherein the rate of cell growth comprises a doubling rate of 10-35 hours.

39. The method of claim 37, wherein the rate of cell growth comprises a doubling rate of 15-30 hours.

40. The method of claim 37, wherein rate of cell growth comprises a doubling rate of 18-26 hours.

41. The method of claim 25, wherein the doubling time comprises conditions such that cell viability is greater than 90%.

42. The method of claim 25, wherein the culture used to prepare the high titer virus is grown for 3-7 days.

43. The method of claim 25, wherein the culture used to prepare the high titer virus is grown for 4-6 days.

44. The method of claim 25, wherein the culture used to prepare the high titer virus is grown for about 5 days.

45. Insect cells producing recombinant RAGE or a fragment thereof according to claim 25.

46. Recombinant RAGE produced by the method of claim 25.

47. A method for high level expression of recombinant human Receptor for Advanced Glycated Endproducts (RAGE) comprising:

(a) preparing recombinant virus comprising a nucleotide sequence encoding RAGE or a fragment thereof subcloned into the *Autographa californica* nuclear polyhedrosis virus;

(b) infecting insect cells at a multiplicity of infection (MOI) of about 0.1 to 0.2;

(c) incubating the insect cell culture at a temperature ranging from 26-28 °C for 3-7 days to prepare high titer virus stock;

(d) titering the virus to determine MOI;

(e) initiating cultures of insect cells at an initial density of about 2.5×10^5 cells per ml;

(f) growing the insect cells such that the growth rate comprises a doubling time of about 18-26 hours and the cells comprise a viability of greater than 90% until the cell density comprises 1.5 to 2.5 x 10^6 cells per ml;

(g) adding virus (from step (d)) at a MOI of 0.1 to 10 to the insect cells; and

(h) incubating the infected culture at about 26-28 °C for a predetermined time or until cloudy.

48. The method of claim 47, wherein the recombinant RAGE polypeptide or fragment thereof is purified from the insect media using Sepharose.

49. The method of claim 47, further comprising a yield of recombinant protein of at least 25 mg per liter of culture.

50. The method of claim 47, further comprising a yield of recombinant protein of more than 100 mg per liter of culture.

51. The method of claim 47, further comprising a yield of recombinant protein of more than 250 mg per liter of culture.

52. The method of claim 47, wherein the nucleotide sequence encoding RAGE comprises SEQ ID NO: 1, or a sequence substantially homologous thereto.

53. The method of claim 47, wherein the fragment of RAGE is the soluble, extracellular portion of RAGE (sRAGE), as defined by the nucleic acid sequence SEQ ID NO: 2, or a sequence substantially homologous thereto.

54. The method of claim 47, wherein the fragment of RAGE is the V-domain of RAGE, as defined by the nucleic acid sequence SEQ ID NO: 4, or a sequence substantially homologous thereto.

55. Insect cells producing recombinant RAGE or a fragment thereof according to claim 47.

56. Recombinant RAGE produced by the method of claim 47.

57. A method of treating human disease comprising administering the recombinant RAGE polypeptide of claim 1 in a pharmaceutically acceptable carrier.

58. The method of claim 57, wherein the human diseases treated using the recombinant RAGE polypeptide comprise atherosclerosis, diabetes, symptoms of diabetes late complications, amyloidosis, Alzheimer's Disease, cancer, inflammation, kidney failure, systemic lupus nephritis, inflammatory lupus nepritis, or erectile dysfunction.

59. A method for inhibiting the interaction of an advanced glycosylation end products (AGEs) with RAGE in a subject, comprising administering to the subject a therapeutically effective amount of recombinant RAGE polypeptide of claim 1.

60. The method of claim 59, wherein the recombinant RAGE is administered as a therapeutically effective amount of recombinant sRAGE with an appropriate pharmaceutical so as to prevent or ameliorate disease associated with increased levels of advanced glycosylation end products (AGEs).

61. The method of claim 60, wherein the disease associated with increased levels of AGEs comprises accelerated atherosclerosis, diabetes, Alzheimer's Disease, inflammation, systemic lupus nephritis, inflammatory lupus nephritis, cancer, or erectile dysfunction.

62. The method of claim 60, wherein the effective amount of sRAGE ranges from about 1 ng/kg body weight to 100 mg/kg body weight.

63. The method of claim 60, wherein the effective amount of sRAGE ranges from about 1 μg/kg body weight to 50 mg/kg body weight.

64. The method of claim 60, wherein the effective amount of sRAGE ranges from about 10 μg/kg body weight to 10 mg/kg body weight.